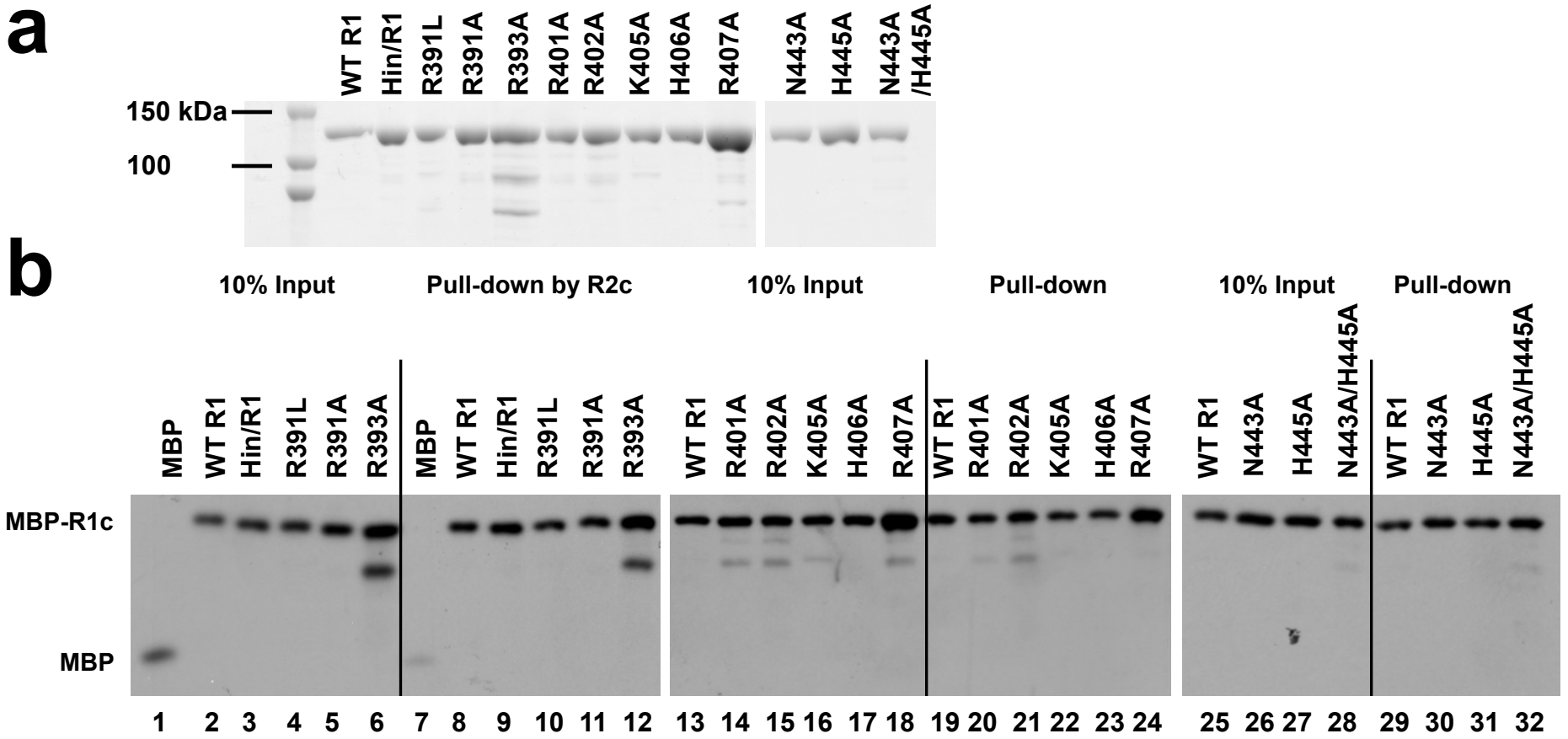

Supplementary Figure 4 GST-RAG2 pull-down of WT and mutant RAG1 core proteins. (**a**) WT and mutant RAG1 core proteins (expected size 110 kDa) were purified from bacteria and analyzed by SDS-PAGE followed by staining with Coomassie Blue. (**b**) Purified GST-RAG2 core and MBP-RAG1 core proteins were incubated together and pulled down using glutathione beads. Western blots of the pull-down samples were probed with anti-MBP antibody to visualize the amount of MBP-RAG1 core protein in each sample. 10% of each input was loaded as positive controls. Reaction in lane 7 contained the MBP tag alone with GST-RAG2 and serves as a negative control.

Method: 1 μg of each MBP-RAG1 and GST-RAG2 core proteins were incubated in 20 μL binding reactions containing 30 mM Tris pH 7.5, 5 mM $MgCl_2$, 100 mM NaCl, and 5% (v/v) glycerol for about 30 minutes on ice before adding 500 μL of the same reaction buffer and 30 μL of pre-washed glutathione beads (GE Healthcare) and incubating on ice for an additional 1 hour. The beads were washed with the same binding buffer 5 times before resuspending in loading dye. The samples were boiled and electrophoresed on denaturing SDS-polyacrylamide gels followed by Western analysis. Western blots were probed with anti-MBP antibody (New England Biolabs) to visualize the amount of MBP-RAG1 pulled down by RAG2.